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# Release calls of four species of Phyllomedusidae (Amphibia, Anura)

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## Abstract

Anurans emit a variety of acoustic signals in different behavioral contexts during the breeding season. The release call is a signal produced by the frog when it is inappropriately clasped by another frog. In the family Phyllomedusidae, this call type is known only for *Pithecophus ayeaye*. Here we describe the release call of four species: *Phyllomedusa bahiana*, *P. sauvagii*, *Pithecopus rohdei,* and *P. nordestinus*, based on recordings in the field. The release calls of these four species consist of a multipulsed note. Smaller species of the *Pithecopus* genus (*P. ayeaye*, *P. rohdei* and *P. nordestinus*), presented shorter release calls (0.022–0.070 s), with higher dominant frequency on average (1508.8–1651.8 Hz), when compared to the bigger *Phyllomedusa* (*P. bahiana* and *P. sauvagii*) (0.062–0.107 s; 798.7–1071.4 Hz). For phyllomedusid species, the release call might indicate a phylogenetic signal, because species of the same genus have similar acoustic traits.

## Key Words

vocalization, animal behavior, bioacoustics, *Phyllomedusa*, *Pithecopus*

## Introduction

The complexity of anuran calling behaviour has been revealed for several Neotropical species (Pombal et al. 1994; Bastos and Haddad 1995; Martins et al. 1998). Anurans emit a variety of acoustic signals in different behavioral contexts during the breeding season (Cardoso and Haddad 1984; Martins and Haddad 1988; Bastos and Haddad 1995, 2002). Besides advertisement calls, some species frequently emit courtship, encounter, reciprocal, territorial, and release calls (Given 1987; Bastos and Haddad 2002).

The release call is a signal produced by the frog when it is inappropriately clasped by another frog (Duellman and Trueb 1986; Wells 2007; Gollmann et al. 2009; Toledo et al. 2014). There are also males that emit release calls during physically aggressive interactions (Bastos and Haddad 1995; Brunetti et al. 2015). Release calls are expected to have less selective pressures than advertisement calls (Brown and Littlejohn 1972), and for some cases of

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cryptic speciation, this call type is a valuable taxonomic tool (Castellano et al. 2002; Grenat and Martino 2013).

The family Phyllomedusidae is currently composed of 65 species distributed in eight genera (Frost 2018), and the release call is known only for *Pithecopus ayeaye* B. Lutz, 1966 (Nali et al. 2015). In addition, Wogel et al. (2004), describing the vocal repertoire of *Pithecopus rohdei* (Mertens 1926), defined the release call of this species as similar to its territorial call type 1 with lower intensity. However, the authors have not formally described the release call of *P. rohdei*.

Given the importance of the release call for anuran systematic, and the few data about this call type for phyllomedusids, herein we describe the release call of four species: *Phyllomedusa bahiana* A. Lutz 1925, *P. sauvagii* Boulenger 1882, *Pithecopus rohdei*, and *P. nordestinus* (Caramaschi 2006).

## Methods

We use acoustic data from different field works. All recorded calls were emitted in the field when the males were being handled, pressed in both sides of the body, in the axillary region, simulating an amplexus.

We summarize all data on the release calls recorded in Table 1. We analyzed all the recordings in the software Raven Pro 1.5 for Mac (Bioacustic Research Program 2012), with the following settings: window type = Hann, window size = 256 samples, 3dB filter bandwidth = 270 Hz, brightness = 55%, contrast = 55%, overlap = 50%, DFT size = 256 samples, grid spacing = 188 Hz. We analyzed acoustic parameters typically evaluated in anuran taxonomy studies: call duration, pulse number per call, pulse rate (pulses per second), and dominant frequency (we used 'Peak Frequency Hz' automatic function of the Raven, by manual selection of call units). Terminology of call descriptions follows Köhler et al. (2017). We constructed audio spectrograms in R using the package *seewave* (Sueur et al. 2008) with the following parameters: FFT window width = 256, Frame = 100, Overlap = 75, and flat top filter.

Comparative data for other species were obtained from the available literature (see Wogel et al. 2004; Nali et al. 2015) (Table 2). We deposited the sound files in the Fonoteca Mapinguari da Universidade Federal de Mato Gros-

**Table 1.** Data on the release calls recorded on the species of *Phyllomedusa* and *Pithecopus* analyzed in this study.

| Species | Locality | Coordinates | Altitude (m) | Recorder | Date | Air Temperature (°C) | Individual Label/Record |
|---|---|---|---|---|---|---|---|
| *Phyllomedusa bahiana* | Serra de São José, Feira de Santana, Bahia | 12°5'45.04"S, 39°2'47.49"W | 350 | Marantz PMD660 | 8 Feb 2012 | NA | MZFS3934/ MAP-V0205 |
| *Phyllomedusa bahiana* | Serra de São José, Feira de Santana, Bahia | 12°5'45.04"S, 39°2'47.49"W | 350 | Marantz PMD660 | 8 Feb 2012 | NA | MZFS3935/ MAP-V0206 |
| *Phyllomedusa sauvagii* | Estância Mimosa, Bonito, Mato Grosso do Sul | 20°58'49"S, 56°30'32"W | 450 | Tascam DR-40 | 8 Jan 2017 | 27.0 | ZUFMS-AMP10561/ MAP-V0209 |
| *Pithecopus rohdei* | Barão de Monte Alto, Minas Gerais | 21°16'29.30"S, 42°14'5.40"W | 450 | Olympus DM-420 | 21 Aug 2013 | 30.9 | MZUFV 13898/ MAP-V0208 |
| *Pithecopus nordestinus* | São Bento do Norte, Rio Grande do Norte | 5°6'34"S, 35°55'50"W | 10 | Panasonic RR-US450 | 29 Mar 2012 | NA | Unvouchered/ MAP-V0207 |

**Table 2.** Release call of *Phyllomedusa bahiana*, *Phyllomedusa sauvagii*, *Pithecopus nordestinus*, *Pithecopus rohdei*, and *Pithecopus ayeaye*. Values are presented as mean, ± SD (range), SD = standard deviation. SVL = snout vent-length.

| Genus | Taxa/Acoustic parameters | SVL (mm) | *n* (calls) | Duration (s) | Pulses/call | Pulses/s | Dominant Frequency (Hz) | Reference |
|---|---|---|---|---|---|---|---|---|
| *Phyllomedusa* | *P. bahiana* male 1 | 76.62 | 35 | 0.075 ± 0.005 (0.063–0.087) | 6 ± 0.37 (5–7) | 81 ± 4.25 (74–90) | 1071.43 ± 201.18 (750.00–1875.00) | This study |
| | *P. bahiana* male 2 | 72.74 | 18 | 0.073 ± 0.009 (0.062–0.096) | 6 ± 0.49 (5–7) | 83 ± 6.48 (66–92) | 895.83 ± 188.11 (562.50–1312.50) | This study |
| | *P. sauvagii* | 77.95 | 33 | 0.087 ± 0.008 (0.078–0.107) | 6 ± 0.44 (6–7) | 72 ± 3.66 (61–78) | 798.67 ± 127.87 (516.80–1033.60) | This study |
| *Pithecopus* | *P. nordestinus* | – | 29 | 0.036 ± 0.008 (0.022–0.053) | 6 ± 0.86 (4–7) | 158 ± 18.06 (113–194) | 1508.81 ±187.92 (1378.10–2239.50) | This study |
| | *P. rohdei* | 39.30 | 18 | 0.043 ± 0.009 (0.034–0.070) | 4 ± 0.47 (3–5) | 92 ± 12.09 (71–118) | 1550.40 ± 350.82 (861. 30–1894.90) | This study |
| | *P. ayeaye* | 31.28–36.60 | 113 | 0.051 ± 0.008 (0.031–0.072) | 9.5 ± 2.70 (5–17) | – | 1651.80 ± 612.90 (861.30–4651.20) | Nali et al. (2015) |

so do Sul, and the voucher specimens for call recordings are housed in Museu de Zoologia de Feira de Santana, Universidade Estadual de Feira de Santana, Bahia state, Museu de Zoologia João Moojen da Universidade Federal de Viçosa, Minas Gerais state, and Coleção Zoológica de Referência da Universidade Federal de Mato Grosso do Sul, Mato Grosso do Sul state (see Table 1).

## Results

The vocalizations were emitted in call series with irregular intervals between calls, while we stimulated the individual. The release call of *Phyllomedusa bahiana* had an ascendant amplitude modulation and slight frequency modulation, in which the band of frequency is narrower at the beginning and broader at the end. The call consists of a single pulsed note, with duration of 0.062–0.096 s, and dominant frequency of 562.50–1875.00 Hz. The number of pulses per call was 5–7, and the number of pulses per second (pulse/rate) was 66–92. The release call of *P. sauvagii* had an ascendant amplitude and frequency modulation on the first half of the call, and descendant in the end. The call consists of a single pulsed note, with duration of 0.078–0.107 s, and dominant frequency of 516.80–1033.60 Hz. The number of pulses per call was 6–7, and the number of pulses per second (pulse/rate) was 61–78. The release call of *P. nordestinus* had no amplitude and frequency modulation. The call consists of a single pulsed note, with duration of 0.022–0.053 s, and dominant frequency of 1378.10–2239.50 Hz. The number of pulses per call was 4–7, and the number of pulses per second (pulse/rate) was 113–194. The release call of *P. rohdei* had an ascendant amplitude and frequency modulation on the first half of the call, and descendant in the end, and had no frequency modulation. The call consists of a single pulsed note, with duration of 0.034–0.070 s, and dominant frequency of 861.30–1894.90 Hz. The number of pulses per call was 3–5, and the number of pulses per second (pulse/rate) was 71–118 (Fig. 1).

Smaller species of the *Pithecopus* genus (*P. ayeaye*, *P. rohdei* and *P. nordestinus*), presented shorter release calls (0.022–0.070 s), with higher dominant frequency on average (1508.8–1651.8 Hz), when compared to the bigger species of the genus *Phyllomedusa* (*P. bahiana* and *P. sauvagii*) (0.062–0.107 s; 798.7–1071.4 Hz) (Table 2). The species with the highest pulse rate is *Pithecopus nordestinus* (113–194 pulses/s), and the number of pulses per note overlaps among species.

## Discussion

Despite none release call of a phyllomedusid frog had been described (besides *P. ayeaye*), maybe this call type was confused with other distress calls. For example, the so-called "Territorial call II" described by Vilaça et al. (2011) for *P. nordestinus*, overlaps all parameters with the release call of *P. rohdei* and *P. nordestinus* described here. The authors described the call as territorial, and suggested a distress call function, because they were emitted while the individual were being held by the researcher (Vilaça et al. 2011). Contrastingly, distress calls are characterized by loud, explosive calls emitted in response to disturbance or potential predators (Duellman and Trueb 1986), thus all the characteristics described in Vilaça et al. (2011) lead us to conclude that they presented the species release call.

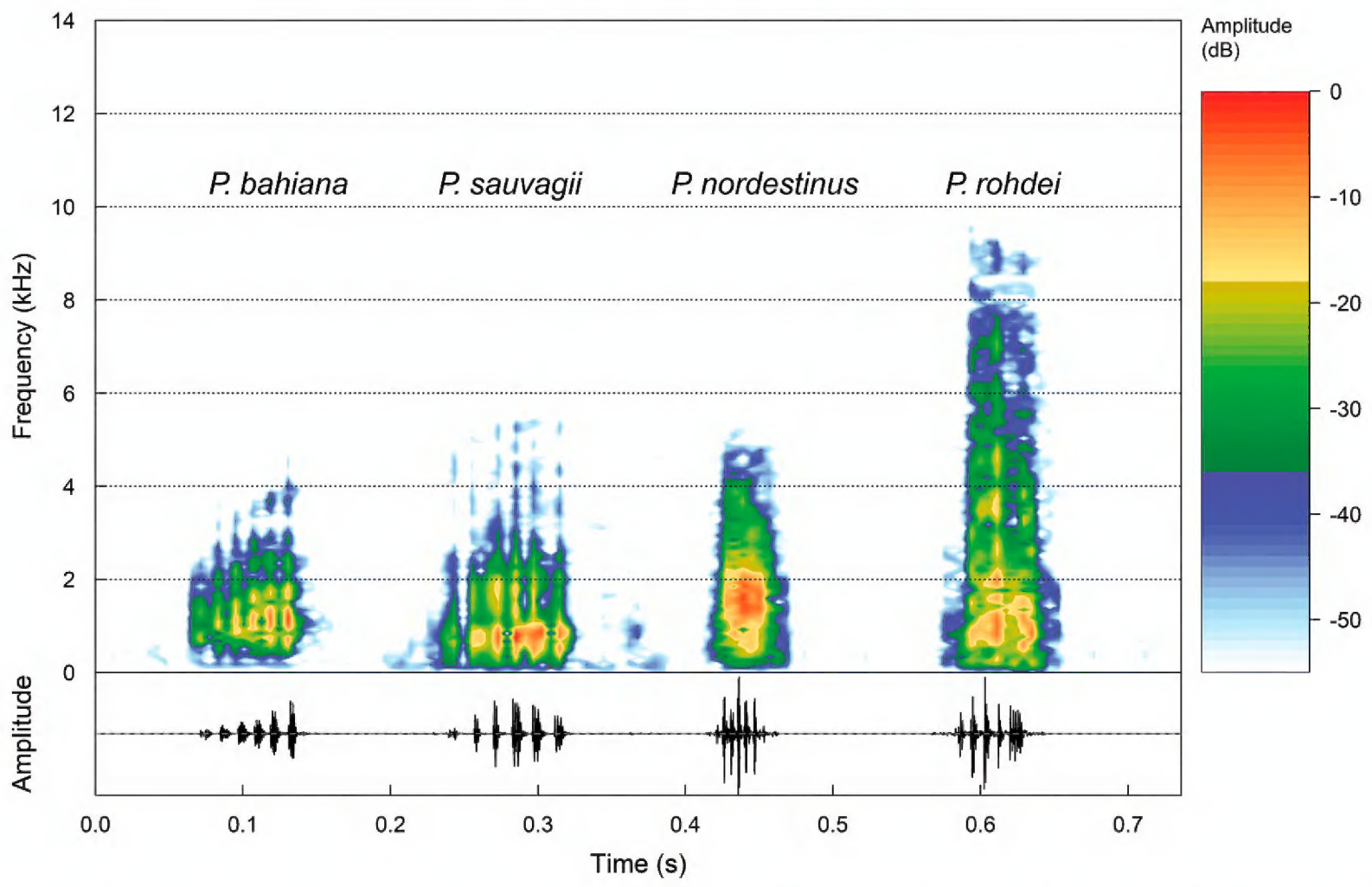


**Figure 1.** Release call of *Phyllomedusa bahiana*, *Phyllomedusa sauvagii*, *Pithecopus nordestinus*, and *Pithecopus rohdei*.

Release calls are emitted by males when they are inappropriately clasped by males, or by females when they are unreceptive, which affirms this type of call as a signal for sex recognition (Wells 2007; Liao and Lu 2009). For phyllomedusid species, the release call might indicate a phylogenetic signal, because it grouped the species that belong to the same genus, based on similarities on the acoustic traits (see Table 2). For other groups of anurans, the release call covaries with genetic distance, suggesting that stochastic processes might have great importance in the evolution of this trait (Castellano et al. 2002).

Although we observed some differences in the release call in different genera within Phyllomedusidae (i.e. *Phyllomedusa* and *Pithecopus*), the calls are similar between species of the same genus (e.g. *Phyllomedusa bahiana* vs. *P. sauvagii*; *Pithecopus nordestinus* vs. *P. rohdei* vs. *P. ayeaye*). A similar pattern was also observed regarding the advertisement calls from others species belonging to the Phyllomedusidae family (e.g. *Phyllomedusa burmeisteri* vs. *P. bahiana*, Andrade et al. 2018; *Pithecopus nordestinus* vs. *P. azureus*, Haga et al. 2017). In these cases, the authors did not find acoustic traits to distinguish the studied species. Nevertheless, the release call differed specifically among closely related species from other groups, and could be phylogenetically informative to distinguish different clades (Brown and Littlejohn 1972; Sullivan and Wagner Jr. 1988; di Tada et al. 2001; Grenat and Martino 2013). In fact, differences in release calls have been used to separate species within *Rhinella spinulosus* group (di Tada et al. 2001), and between *Odontophrynus americanus* and *O. cordobae* (Grenat and Martino 2013).

The description of release calls of other species of the family Phyllomedusidae is highly important, which combined with the advertisement call, can reveal its utility for phyllomedusid systematics, within the family and its groups. Besides, such descriptions provide data that can be useful in studies on diversity and evolution of phenotypic traits or even how such signals are used in inter and intra-specific interactions.

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